Influence of fines on commercial lactose carriers and their dry powder inhalation performance

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Summary

Many efforts have been made in the past to explain the beneficial effect of fines but the exact mechanisms how these fine lactose particles alter the performance of dry powder inhaler (DPI) formulations has remained unclear. In this study the influence of fines added to commercially available lactose carriers is investigated. For this purpose, two different-sized carriers were blended with different concentrations of fines and the model drug budesonide in different blending orders. All blends were prepared with a high shear mixer, which would make an upscaling possible, at different rotational speeds and mixing times. Afterwards the blends were tested with an inhaler device (Novolizer®) by using the Next Generation Pharmaceutical Impactor (NGI). The fine particle fraction (FPF), the fine particle dose (FPD) and the mass median aerodynamic diameter (MMAD) were then compared with the factors rotation speed, blending time, blending order and the amount of fines. It could be shown that all factors have an impact on the FPF and FPD. As an explanation all common postulated theories come into consideration. With the help of blending order results and SEM pictures the saturation of active sites, the function of the fines as a buffer during blending and the formation of drug-fines agglomerate could be proven. Overall, the great benefit of fines for commercial lactose carrier and their DPI performance could be shown in this work as well as the importance of a better understanding of the exact mechanism of their acting, in particular for the blending process.

Introduction

The addition of lactose fines (< 15 µm) in ternary powder mixtures for inhalation is well-known to increase the fine particle fraction (FPF, < 5 µm) but the exact mechanisms for how these fines work, remain unclear. Numerous theories like the active sites hypothesis[1], agglomeration hypothesis[1], fluidisation hypothesis[2] and the buffer hypothesis[3] have been discussed. The active sites hypothesis suggests that the fines occupy areas of the surface which are more adhesive, leaving only weaker binding sites available for the drug particles to bind to. The agglomeration hypothesis proposes that fines form agglomerates, multiplets or multi-layers with drug particles, which are supposed to be aerosolised more easily than single drug particles. Focusing on this theory Kinnunen et al. exhibited a co-deposition of drug-fines agglomerates and an increase of the mass median aerodynamic diameter **(**MMAD) with higher fine contents[4]. The fluidisation hypothesis suggests that fines increase the tensile strength of a bulk powder, which increases the minimum energy required for fluidisation during inhalation. This effect, combined with greater frequency of particle-particle and particle-wall collisions, results in a better dispersion performance. Grasmeijer et al. exhibited that fines coarser than the drug particles act as a buffer between colliding carrier particles and protect drug particles from press-on forces during blending. He further suggested that all of these above-mentioned theories act simultaneously[3]. In this study, the impact of these mechanisms on commercially available lactose grades was tested. For this purpose, the influences of rotation speed, blending time and blending order on binary and ternary powder mixtures were investigated with a high shear mixer. In addition, the influences of fines on FPD/FPF were tested with a commercial inhaler device (Novolizer®).

Material and methods

Preparation of model DPI formulations: An Alpine Picoline equipped with the Picomix® high shear mixer module (Hosokawa Alpine, Augsburg, Germany) was used to prepare adhesive mixtures with InhaLac 70® and InhaLac 230® as a carrier, InhaLac 400® as fines (all Molkerei Meggle Wasserburg GmbH & Co. KG, Wasserburg, Germany) and micronised budesonide (Farmabios, Gropello Cairoli, Italy) as a model drug. Budesonide concentration was set to 1.5 wt% to get a delivered dose of approximately 200 µg with the Novolizer® (Astellas Pharma GmbH, München, Germany). 2.5, 5.0 or 7.5 wt% of fines were added to the blends in five different blending orders (Table 1). The blending time for every blending step was varied between 30 and 300 s to reach total blending times of 60 to 600 s. The rotation speed was altered between 500 and 1500 rpm. Budesonide and InhaLac 400® were sieved with a 180 µm sieve prior blending to remove agglomerates. A sandwich-weighing-method was used at a batch size of 55 g for blending. After every blending step, the mixtures were passed through a 355 µm sieve to remove agglomerates. Homogeneity was assessed for all formulations by measuring the budesonide content of ten randomly drawn samples by an HPLC method. The requirements were set to a relative standard deviation (RSD) of less than 5 % and a recovery of 95 %. Each formulation was stored at room conditions for a minimum of one week before NGI analysis.

Table 1 – Preparation details for the various formulations tested. C = carrier, API = budesonide and F = fines.

Formulation	First blend	Second blend	Rotation speed [rpm]	Total blending time [s]
[C + API]	C + API	[C + API]	500	120
[C + API] + [F]	C + API	[C + API] + F	500, 1000, 1500	60, 120, 240, 600
[C + F] + [API]	C + F	[C + F] + API	500, 1000, 1500	60, 120, 240, 600
[F + API] +[C]	F + API	[F + API] + C	500	120
[C + F + API]	C + F + API	[C + F + API]	500	120

Determination of in vitro fine particle delivery: Aerodynamic particle size distribution was determined by cascade impaction with the Novolizer®. A Next Generation Pharmaceutical Impactor equipped with a TPK trigger box (both Copley Scientific, Nottingham, UK) and a vacuum pump (Erweka, Heusenstamm, Germany) was used. The air flow was set to 78.3 L/min as measured with a flow meter (DFM2, Copley Scientific, Nottingham, UK) resulting in a pressure drop of 4 kPa across the Novolizer®. Afterwards the air flow was applied over 3.1 s to draw 4 L of air through the mouthpiece of the inhaler. A stage coating (Brij®35, glycerol, absolute ethanol) was applied onto the preseparator and each stage of the impactor before analysis. 10 actuations were applied to the NGI for one run. Induction port (including adapter), preseparator and all stages were individually sampled for budesonide. Every blend was measured in triplicate. Calculation of FPD, FPF and MMAD was done with the software CITDAS 3.0 (Copley Scientific, Nottingham, UK). NGI results were evaluated with Modde® (version 10.0, Umetrics AB, Umea, Sweden) by using the multiple linear regression (MLR) method. Rotation speed, blending time, content of fines and the blending order were chosen as factors. The model was evaluated by neglecting insignificant terms in a backward regression and the FPF and FPD were chosen as response for this study.

Particle size analysis: Laser light diffraction was performed to analyse the particle size distribution (PSD). Samples were dispersed with 3 bar using a Sympatec HELOS laser diffractometer with the RODOS dry dispersion system (Sympatec GmbH, Clausthal-Zellerfeld, Germany) and a suitable lens. The powder was fed into the dispersing system by using a spatula. Data acquisition and calculation was performed with Windox software 5.4.2.0. (Sympatec GmbH, Clausthal-Zellerfeld, Germany) using the Fraunhofer theory. Every sample was evaluated volume based and in triplicate.

Scanning electron microscopy (SEM): The morphology of the substances and blends was examined using SEM. The powder was fixed on a carbon sticker and coated with gold by using a BAL-Tec SCP 050 Sputter Coater (Leica Instuments, Wetzlar, Germany). The samples were then visualized with a Zeiss Ultra 55 plus (Carl Zeiss NTS GmbH, Oberkochen, Germany) using the SE-2 detector and a working voltage of 2kV.

Picomix® performance data: The Picomix® tracks the product temperature, rotation speed and motor output. To determine the motor output 55 g of powder were mixed at different rotation speeds.

Results and discussion

Particle size analysis: The median particle size of InhaLac 70® (Table 2) is around 100 µm and with this larger than InhaLac 230® and it does not contain any intrinsic fines. InhaLac 70® shows no change in PSD at a blending speed of 500 rpm, but the particles get smaller for higher rotation speeds. This can be explained by the high energy input during blending (Figure 1). InhaLac 230® is much smaller and more stable due to the smaller size leading to no reduction of the PSD with increasing rotation speeds. InhaLac 400® shows larger particles and a broader PSD than budesonide. This is very important for buffer effects during blending, because fine lactose particles being coarser than the drug particles can act as a buffer between colliding carrier particles and protect the API from press-on forces.

Table 2 - Particle size distribution of the mixture components (average (SD); n = 3)

Material	d_{10} [µm] ± SD			d_{50} [µm] ± SD			d_{90} [µm] ± SD		
InhaLac 70	126.9	±	0.6	218.7	±	1.0	302.4	±	1.3
InhaLac 70 *	125.6	±	0.7	218.5	±	1.6	303.6	±	1.1
InhaLac 70 **	105.8	±	1.4	208.9	±	1.7	298.8	±	0.8
InhaLac 230	55.7	±	0.1	101.3	±	0.1	142.6	±	0.1
InhaLac 230 *	56.3	±	0.1	101.2	±	0.2	142.2	±	0.1
InhaLac 230 **	57.3	±	0.1	102.2	±	0.2	143.1	±	0.3
InhaLac 400	1.0	±	0.0	6.3	±	0.0	23.9	±	0.4
Budesonide	0.4	±	0.0	1.5	±	0.0	3.5	±	0.0

* 55 g blended with 500 rpm for 120 s, ** 55 g blended with 1500 rpm for 120 s

Picomix® performance data: As illustrated in figure 1, higher energy consumption is measured when blending InhaLac 70® with the given rotation speeds. An explanation for this can be given by the higher mass of the individual particles and hence their higher inertia. This allows a conclusion about the present press-on forces, which occur to be higher for InhaLac 70® blends than for InhaLac 230®. Intrinsic fines of InhaLac 230 may also act as a "lubricant" and facilitate particle flow and by this reduce the force needed to move the blade.

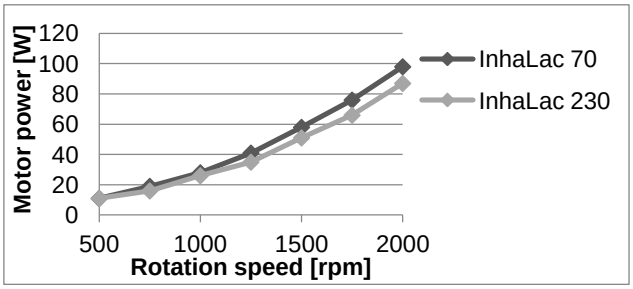

Figure 1- Picomix® performance data with 55 g batches of InhaLac 70® or InhaLac 230®

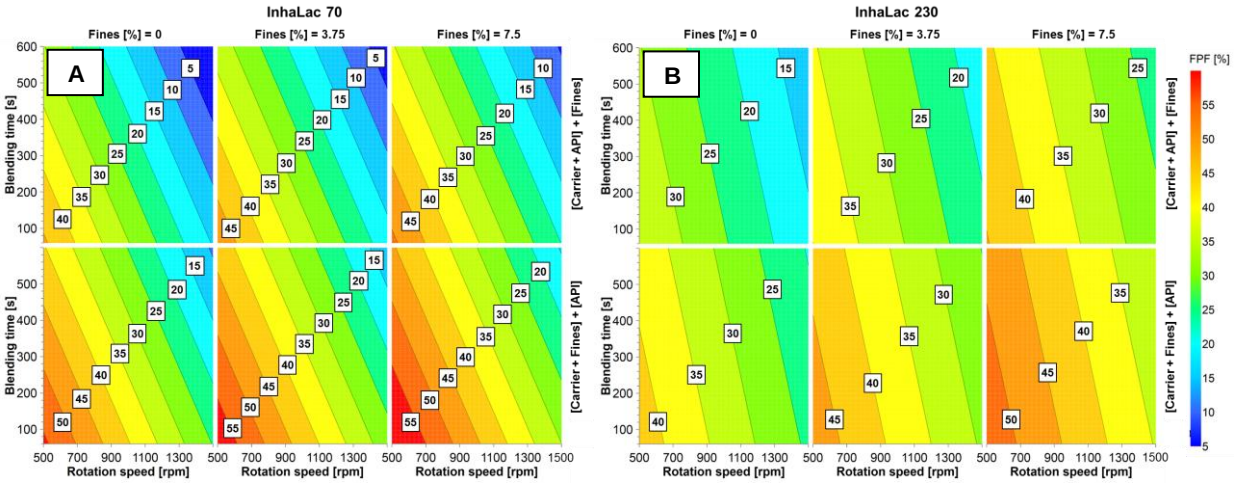

Figure 2 - 4D contour plot of impaction analysis results for InhaLac 70® (**A)** and InhaLac 230® (**B**). Plotted is the mean FPF against the rotation speed, blending time and the content of fines for two different blending orders (upper line: lactose carrier and drug first, then lactose fines, ([C+API]+[F]); lower line: carrier and fines first, then drug, ([C+F]+[API])).

Homogeneity and Recovery: After mixing, all blends with InhaLac 230® show a good homogeneity (all RSD < 3.55 %) and an appropriate recovery (recovery > 95.70 %). This is also the case for most of the blends with InhaLac 70® (RSD < 4.23 %). However, the recovery decreases at high rotation speeds to a minimum recovery of 90.96 % at 1500 rpm. This is due to an increased deposition of drug on the walls and especially the rotor of the blender.

In vitro fine particle delivery for InhaLac 70® (Figure 2A): The quality of the resulting model is good (percent of variation R^2 = 0.95, prediction quality Q^2 = 0.87, reproducibility RP = 0.99, model validity p = -0.09, the poor model validity is a result of an artificial lack of fit). Because of the high reproducibility the pure error is very low and not representative for the true experimental error. That results in significant (but not real) lack of fit. Rotation speed, blending time, content of fines and the blending order are all identified as significant factors on the FPF. The rotation speed is the most important factor on the FPF (-15.58 ± 2.24). Increased rotation speed leads to higher energy input (Figure 1) and an increase of press-on forces, which causes a reduction of the FPF. This reduction is more prominent, when carrier and API were blended first (- 5.08 ± 1.31) and the fines were added in the second blending step. When the fines were added first (5.08 ± 1.31), they can act as a buffer between colliding carrier particles and protect the smaller drug particles from press-on forces. The total effect of fines (2.64 ± 2.11) is not very high. This is due to the fact, that the addition of fines in the second blending step has hardly any beneficial effect. If budesonide was added in the first blending step, it remains in the indentions of the carrier (Figure 3 A and B). No replacement of fines and drug can be observed for prolonged blending times (-7.77 ± 2.60). Opposite, the FPF of the blends decreases considerably with prolonged blending time when the drug was added in the first blending step. The other blending orders do not differ from each other and deliver intermediate high results for the FPF.

An evaluation of the FPD provides comparable results. Interestingly, an increase of MMAD with an increasing content of fines can be determined. For example for the blends with the blending order carrier and fines first, drug added next, the MMAD increases from 1.90 up to 2.15 µm. This phenomenon arises due to a co-deposition of drug-fine agglomerates. These agglomerates can also be seen in the SEM pictures (Figure 3A).

In vitro fine particle delivery for InhaLac 230® (Figure 2B): The quality of the resulting model is just as well as for InhaLac 70® (percent of variation $R^2 = 0.92$, prediction quality $Q^2 = 0.81$, reproducibility RP = 0.98, model validity p = 0.21, poor validity is again a result of an artificial lack of fit). Rotation speed, blending time, content of fines and the blending order is all identified as significant factors on the FPF. The rotation speed has less impact (-9.28 ± 1.74) on the FPF, than for InhaLac 70®. This can be explained by the lower particle size of InhaLac 230® and therefore the lower press-on forces and buffer effect during blending. The blending time (-2.27 ± 2.01) has only a very small but still significant influence on the FPF. The blending orders show similar results compared to InhaLac 70® (3.98 ± 1.02 and 3.98 ± 1.02, respectively) and there is still an effect of fines (5.27 ± 1.64). As an explanation the same mechanisms as mentioned above for InhaLac 70® can be used. FPD and MMAD results correlate to the results of InhaLac 70®. The highest FPF that can be achieved for InhaLac 230® is lower than for InhaLac 70®. That can be explained with the dispersion mechanism of the Novolizer®. The Novolizer® is equipped with an air classifier technology, which is primary based on inertial de-agglomeration forces [5]. The higher mass of the InhaLac 70® particles leads to a better dispersion during inhalation and thus to higher FPFs.

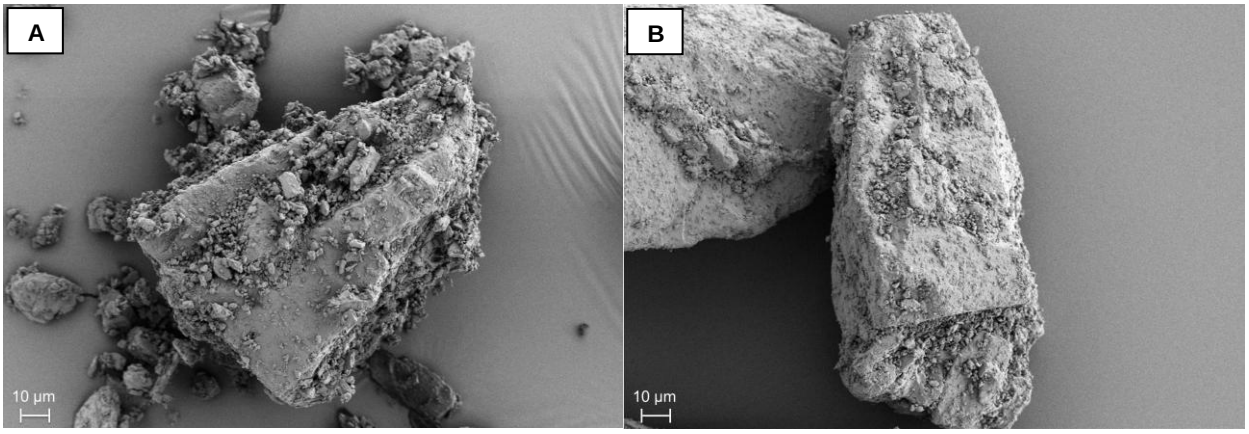

Figure 3 – Visualisation of representative interactive mixtures with (**A**): InhaLac 230 blended with API first, then blended with fines (7.5 %) before dispersion and (**B**): after dispersion with the Novolizer®

Scanning electron microscopy: SEM image (Figure 3A) shows high loading of the carrier surface with agglomerates of drug particles and fines. Most particles are attached to the indented parts of the carrier whereas the smoother parts show less loading. After dispersion it is more evident that particles are deposited preferentially in the indentions of the carrier (Figure **B**). API and fines remain in the indentions supporting the active sites theory and giving an explanation for blending order results.

Conclusion

Because of the saturation of active sites and the reduction of press-on forces the correct blending order is an important factor for the production of powder blends for inhalation with InhaLac 70® and InhaLac 230® as carrier and InhaLac 400® as fines. Other mechanisms like the formation of agglomerates of fines and drug and the increased tensile-strength also play an important role. Blending time and rotation speed are significant factors, but depend on the used carrier. The larger carrier InhaLac 70® shows greater dependence on the rotation speed and blending time than the smaller InhaLac 230® and is able to achieve higher FPF with the Novolizer® as it is the case for InhaLac 230®.

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